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TITLE: A Phase II Immunotherapeutic Trial: Combination Androgen Ablative Therapy Treatment for Advanced Prostate Cancer

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14. ABSTRACT:
The objectives of this study are to test whether CTLA 4 blockade + AA therapy canenhance clinical treatment responses in advanced prostate cancer patients relative to AA therapy alone. Study patients are randomized to 3 months of combined AA therapy + MDX-010 or versus AA therapy alone. To date, 46 patients have been enrolled and 42 randomized per protocol. An additional 84 patients have been screened and deemed ineligible for study. 29 patients now have sufficient follow-up to assess if any treatment effects may be occurring. In general, patients receiving combined AA + MDX-010 have exhibited greater PSA responses than patients receiving AA therapy alone. We have also observed that MDX-010 does not likely affect initial testosterone production but may delay testosterone recovery. Our preliminary studies also indicate atypical and favorable responses to combined MDX-010 + AA therapy including reversal of rising PSA, rapid resolution of obstructive urinary pathology and dramatic tumor downstaging resulting in unexpected 1 year disease-free patient status. Based on these preliminary observations, we believe that combined AA + MDX-010 treatment may encompass a promising approach to improve advanced prostate cancer treatment.

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I. Introduction

Based on the function of the T cell CTLA-4 receptor, it is anticipated that in vivo CTLA-4 blockade (using MDX-010) will enhance responses to androgen ablative (AA) therapy by boosting prostate-specific and generalized host T cell activity in prostate cancer patients. Such potentiation of host T cell activity by CTLA-4 blockade might, in turn, correlate with superior clinical responses for patients receiving MDX-010 antibody + AA therapy (relative to patients initially receiving AA therapy alone). To test this, we are conducting this open-label randomized phase II trial in which 108 patients with advanced prostate cancer are being prospectively enrolled onto study. Upon enrollment, patients are immediately randomized to receive either: **A)** Three months of androgen ablative (AA) therapy plus a single intravenous infusion of 3 mg/kg MDX-010 (treatment group) versus **B)** three months of AA therapy alone (control group) followed by the option for a single infusion of intravenous 3 mg/kg MDX-010; given upon PSA progression to greater than 4.0 ng/mL following cessation of AA therapy. Equal numbers of control and treatment group patients are to be enrolled onto study.

- The primary study endpoint is PSA progression to > 4.0 ng/mL after cessation of AA therapy. The primary endpoint for this trial will be the proportion of patients remaining progression-free. The progression free interval is defined as the interval from the start of AA therapy until PSA progression, i.e., a rise in PSA to >4.0 ng/mL demonstrated twice in measurements taken two weeks apart.

- The secondary endpoint for study includes parameters of PSA response to treatment such as absolute nadir values, time-until-nadir and rates of decline in PSA.

- The tertiary endpoints for this study are patient immunologic indices of anti-tumoral response and enhanced T cell activation.

- As a cross-over component of this trial, control patients receive CTLA-4 blockade (MDX-010) upon disease progression. PSA response, in this case, is defined by a reduction in PSA of ≥ 50% (demonstrated twice in measurements taken two weeks apart) following the administration of MDX-010 compared with the PSA obtained just prior to MDX-010 administration. Disease progression for the cross-over group is defined as a rise in PSA following MDX-010 administration. Patients demonstrating insufficient changes in PSA to qualify for either response or progression are considered to have *stable* disease.

II. Partial Synopsis of Grant and Protocol History and Changes

• Notification of funding for the scientific application which serves as the framework for this clinical protocol was originally given by the DOD CMPCRP to Eugene D. Kwon MD in 2000. At that time, Dr. Kwon was an investigator and attending physician at the Loyola University Medical Center in Maywood, Illinois. Key preclinical trial scientific

observations that provided compelling rationale for this study included: 1) the published observation that CTLA-4 blockade can be used to potentiate antitumoral immunity; 2) published observations that in vivo CTLA-4 blockade in mice can facilitate murine prostate tumor regression; 3) the published observation that CTLA-4 blockade may require a tumor-specific vaccination scheme in order to enhance specific responses directed against the cancer to be targeted; 4) the published observation that AA therapy in humans prompts prostate tumor infiltration by T cells that might further be amenable to potentiation by CTLA-4 blockade resulting in accelerated prostate cancer regression due to enhanced immunotherapeutic activity directed against prostate tumor cells; 5) the recent availability of a fully humanized, commercially-developed, monoclonal anti-CTLA-4 antibody (MDX-010; Ipilimumab) that had completed phase I trial testing as a reagent for the potential treatment of prostate cancer as well as melanoma.

• Following DOD notification to fund the scientific application submitted by Dr. Kwon, the DOD subsequently requested that a clinical protocol be fully developed in order to execute the aims as outlined in the DOD-funded scientific proposal. As such, between 2000 and 2004, substantive time and attention were directed towards the generation of a clinical protocol entitled "A Phase II Immunotherapeutic Trial: Combination Androgen Ablative Therapy and CTLA-4 Blockade as a Treatment for Advanced Prostate Cancer." Included in this initial effort was: a) the conceptualization and writing of a phase II clinical trial protocol; b) generation of case report forms; c) extensive input into study design by two study statisticians; d) the filing of an investigator IND with the FDA; e) procurement of local IRB approvals; f) procurement of FDA-approval to conduct study; g) negotiation and coordination with the University of California, San Francisco (co-Investigator, Dr. Eric Small) to serve as a sister site to execute a two-institution, phase II clinical trial to support patient accrual and treatment for the proposed study; h) negotiating an agreement with Medarex to provide free-of-charge study drug; i) legal negotiation of language between the study institution and the DOD regarding indemnification and protection of patients to be enrolled onto study; and j) procurement of USAMRMC HSRRB approval to conduct the study.

• Following partial completion of the above cited activities, Dr. Kwon moved to the Mayo Clinic in Rochester Minnesota in August of 2002. Dr. Mishra of the DOD immediately facilitated transfer of grant activities and funds from Loyola to the Mayo Clinic, and Loyola responded by relinquishing the grant and funding to Mayo. At this time, negotiations pertaining to patient indemnification consent language were still undergoing consideration by the DOD. Loyola's IRB had granted IRB approval. IRB approval by Mayo, UCSF and the USAMRMC HSRRB were still pending, partly contingent upon finalization of the study consent language including issues pertaining to patient indemnification. In addition, the phase II protocol needed to be re-formatted to conform to Mayo standards and templates and underwent re-review by the Mayo Department of Urology, the Mayo Comprehensive Cancer Center RAS Committee, the Mayo Comprehensive Cancer Center DSMB, the Mayo Comprehensive Cancer Center Statistical Studies Group, Mayo Legal and Mayo's IRB.

• By February 13, 2004, negotiations pertaining to patient indemnification language were concluded between the legal entities representing the DOD and the Mayo Clinic. On this date, the Mayo Clinic received notification from the USAMRAA that Modification No. P00001 for Grant No. DAMD 17-02-1-0245 was fully executed.

• On March 25, 2004, an initiation site visit was conducted by Medarex (in anticipation of opening accrual of patients) at which time it was noted that a minor point regarding drug administration language in the protocol needed to be revised; mandating a change in an IV infusion filter from a 0.22 micron to a 1.2 micron IV filter. Until that change was instituted, drug infusion was no longer feasible in the manner written in the protocol.

• As such, a protocol modification, dated April 8, 2004, was immediately submitted and approved by the Mayo IRB on May 20, 2004. The revised protocol was subsequently submitted to the USAMRMC office. At this time, even though the protocol was considered active and open to enrollment, we were unable to proceed with study enrollment until we could obtain HSRRB approval of the April 8, 2004, amendment, due to the changes in the drug administration language that were required.

• Additional documents were subsequently requested by the HSRRB, and a Memorandum for Record (dated August 18, 2004) was generated in response to the amendments which the USAMRMC office received on June 14, 2004 and the additional information which Dr. Beitins of the HSRRB received on July 12, 2004.

• A site visit from the USAMRMC office took place at Mayo Clinic Rochester on November 19, 2004. The objective of this site visit was to better understand and establish mechanisms to facilitate the opening of this trial for accrual of patients onto study. Multiple discussions, both prior to and during this site visit, centered upon alleviating obstacles associated with opening this study (both at the participating institutions as well as at the DOD). Discussions included accelerating the process of securing IRB and HSRRB approval of amendments and facilitating more rapid opening of accrual at the UCSF site. Members of the USAMRMC (and the HSRRB prior to visit) voiced multiple suggestions including the addition of other study sites to help make up for lost time in order to meet accrual goals. One member of the HSRRB (prior to visit) also made a number of “off the record” recommendations that we consider dramatically broadening eligibility criteria in order to ramp up accrual capabilities. Finally, it was indicated that tertiary (immunologic) endpoints of study may need to be dropped since funds would be insufficient to execute the completion of these “scientific” endpoints.

• On December 20, 2004, the USAMRMC office sent the draft recommendations from the December 8, 2004 HSRRB meeting to Dr. Kwon.

• On January 4, 2005, the official HSRRB review of the protocol (dated December 8, 2004) was sent to Mayo. On January 26, 2005, Mayo responded to the HSRRB meeting requests; a revised protocol and consent dated January 25, 2005 were submitted to the HSRRB. On January 27, 2005, Dr. Kwon received notification from Dr.

Beitins of the HSRRB that the documents submitted on January 26, 2005, were approved by the Acting Chair of the HSRRB. The revised protocol and consent form dated January 25, 2005, were subsequently approved by the Mayo IRB on February 24, 2005. An approval memorandum was issued to the Mayo contract specialist by Colonel Laura Brosch on March 15, 2005. Thus, this relatively simple modification to our protocol (to substitute an IV infusion filter), in essence, introduced a one year delay in the opening of accrual of patients onto this study.

• Recruitment activities commenced at Mayo in early April, 2005, and have been ongoing since that time. During the course of our recruitment activities -- and in response to our Department of Urology faculty (and one of many recommendations related to us by phone by one member of the HSRRB) -- we concluded that the eligibility criteria for our study were too arbitrarily narrow, thereby hampering our overall ability to enroll patients onto study. Research within our own department revealed that approximately 50 percent of the referrals that we received included patients who had recently been started on hormone therapy. We also determined that a significant number of patients, who might otherwise qualify for enrollment onto study, were in fact being excluded due to the "limited metastases" inclusion criterion articulated in our protocol. Therefore, we broadened this specific eligibility criterion (see below) to enhance accrual without, in our estimation, compromising the interpretation of outcome data pertaining to the treatment of patients with local or advanced prostate cancer receiving treatment on the study. In addition, although we anticipated that the UCSF site would enroll 54 participants, we were soon informed that UCSF would be unable to screen or enroll any participants for this trial. Consequently, UCSF requested that they be removed as a study site.

• A protocol revision dated August 28, 2006, was submitted to the Department of Defense on August 31, 2006. Protocol revisions consisted primarily of expanding the inclusion/exclusion criteria to include patients with any T stage prostate cancer, with or without metastatic disease (with the exclusion of central nervous system metastases), staged within 180 days of enrollment, including post-prostatectomy patients with a rising PSA and including patients who have initiated hormone therapy ≤21 days prior to enrollment. Protocol revisions also included removing the University of California, San Francisco as a study site since they had been unable to screen or enroll any participants. The inclusion of post-prostatectomy patients onto study was in response to a member of the DOD HSRRB who suggested that inclusion of such patients might also be useful for study and help ramp up patient accrual. Mayo Urology also concurred that a large number of post-prostatectomy patients were being referred to Mayo with limited options for treatment on study or otherwise.

• These protocol and consent form revisions were approved by the Department of Defense Human Subjects Research Review Board on December 20, 2006, and by the Mayo Cancer Center RAS Committee on December 20, 2006; these revisions were approved by the Mayo IRB on January 11, 2007. These revisions (in particular, the inclusion of patients with extensive metastatic disease and patients who had initiated

hormone therapy ≤21 days prior to enrollment) greatly facilitated the enrollment of patients onto our trial.

• Given that inclusion of patients who had already initiated hormone therapy had such a dramatic effect on improving accrual of patients onto study, another protocol revision dated June 18, 2007, was submitted to the Department of Defense on July 24, 2007. Protocol revisions consisted primarily of further expanding the inclusion/exclusion criteria to include patients who have initiated hormone therapy ≤90 days prior to enrollment since we see a significant number of participants who fall just outside of the ≤21 days of hormone therapy criterion. The intent of this revision was to capture such patients and more than ensure that no further "prior hormone therapy" amendments to our protocol would need to be filed.

• These protocol and consent form revisions were reviewed by the Department of Defense Human Subjects Research Review Board on October 17, 2007. The Board recommended that further revisions to the protocol and consent be made and that additional documents and information be provided. The requested protocol and consent form revisions were made and were submitted to the Mayo IRB. During the course of our IRB review process and after further consideration, our revision to the exclusion criterion expanding the acceptable length of hormone therapy to ≤90 days was subsequently retracted due to the fact that we have recently seen a very rapid increase in our accrual with the current ≤21 days of hormone therapy criterion.

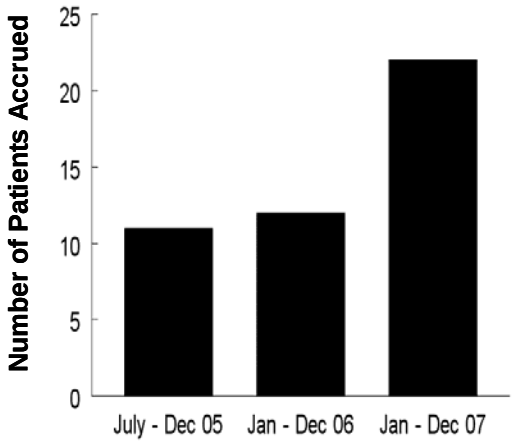
• On December 7, 2007, we received Mayo IRB approval of the 6-18-07 protocol amendment which now basically consists of: 1) clarification only to sections 6.1 and 6.2; 2) editorial changes; 3) updated language in sections 11, 16, 17, 18, and 19 to reflect the current USAMRMC ORP HRPO language and; 4) updates to sections 2.6.1, 2.6.2, 2.6.3, 2.6.5, and 8.10 to include the updated toxicity information on MDX-010, as per the revised Investigator Brochure (Version 10).

• All requested documents and information and the further revised protocol and consent form were forwarded to the HSRRB on December 11, 2007.

III. Clinical Trial Activities

Since the time of our last report, we have enrolled an additional 21 participants to achieve a total of 46 patients consented for study. Of these, 42 have been randomized per protocol at Mayo Clinic Rochester. Four patients who consented were not randomized per protocol for the following reasons: 1) Participant opted for a second opinion closer to home; 2) Total bilirubin was too high to randomize; 3) Participant canceled due to frequent trips to the clinic with his wife who also has cancer; and 4) Amylase was too high to randomize.

Figure 1



Our most recent protocol revisions have dramatically increased our ability to accrue patients onto study as is illustrated in **Figure 1**. Specifically, one major factor that previously excluded candidate patients from study was the injection of Lupron by their referring urologist just prior to seeking second opinion regarding treatment options at the Mayo Clinic. By expanding our eligibility criteria to permit these patients who had initiated hormone therapy ≤21 days prior to their visit to Mayo, we have been able to dramatically increase our enrollment of patients onto study. At present, we are confident that our ability to accrue patients onto trial will continue to increase exponentially.

In addition to patients accrued onto study, we have also had a minimum of 84 formally-documented screen-failures from 5/26/04 through 12/4/07; many of which occurred before the inclusion/exclusion criteria were changed and approved on 2/21/07. Also, innumerable undocumented patients have been deemed ineligible for study by our Urologists and Oncologists. The specific reason for documented screen failures is as follows:

- 14 patients began hormone therapy (before criteria changed)
- 9 patients on hormone therapy >21 days (after criteria changed)
- 12 patients diffuse metastatic disease (before criteria changed)
- 2 patients PSA too low or not rising (to meet entry criteria)
- 11 patients prior treatment (radiation or cryotherapy)
- 1 patient taking Saw Palmetto (before criteria changed)
- 5 patients taking Proscar/Avodart
- 2 patients decided on watchful waiting
- 7 patients decided on surgery despite high risk
- 6 patients post prostatectomy with rising PSA (before criteria changed)
- 1 patient elevated ALT
- 4 patients logistical problems (travel)
- 5 patients with other cancers < 5 years
- 1 patient unable to have biopsy for disease confirmation
- 1 patient refused staging biopsy
- 1 patient significant co-morbidities
- 2 patients refused study due to study risks articulated in the consent.

IV. Continued Efforts to Bolster Accrual of Patients Onto Study.

We continue to work aggressively to accrue participants onto study by maintaining a consistently integrated working relationship with our staff urologists, oncologists, radiation therapists, residents and physician's assistants here at Mayo Clinic. We firmly believe that our efforts are rapidly gaining momentum and garnering substantial support and enthusiasm from our physicians and allied health staff. As such, we plan to continue with the previously established recruitment activities. These activities have consisted of:

- posting study information on the Mayo clinical trials webpage
- posting study information on clinicaltrials.gov
- posting study information on the Mayo Cancer Center Priority Book
- efforts to open the study at Mayo Clinic Arizona and Mayo Clinic Jacksonville
- presenting the study at meetings held locally and nationally
- distributing printed flyers to staff urologists, residents, and physician assistants (PAs)
- posting study flyers on intra-clinic bulletin boards
- posting study flyers in all of the exam rooms
- sending weekly e-mail notifications to staff physicians, residents, and PAs
- scheduling individual one-on-one meetings with Dr. Kwon and the staff urologists
- presentations of the protocol at staff and resident meetings
- daily review of physician calendars for potential participants
- daily telephone calls to physicians, residents, and PAs each morning asking for referrals of any potential participants that they may see during the day
- networking with nurses, technicians, paramedical personnel, appointment schedulers, and other RN study coordinators within the Department of Urology

In addition, we firmly believe that our trial has begun to take on "a life of its own", and awareness for our trial has finally begun to spontaneously filter into, and disseminate within, the national and international arena. Increased interest in our trial has spread by publicizing our study on a national and international basis via word of mouth (patient-to-patient, physician-to-physician, etc) internet postings and informal discussions at national and international venues. In support of this, we have now enrolled patients from Canada, Mexico and the United Arab Emirates as well as distant sites within the United States, including California, Colorado and Missouri. In fact, less than 30% of patients currently accrued onto our study live within our Tri-State region. In addition to this, we routinely screen inquiries to enroll into our study from outside countries including, but not limited to, Sweden, the Middle East and Asia.

V. Measures to Defray Costs Associated with Study

Study patients receive MDX-010 as outpatients in the NCRR-sponsored Clinical Research Unit (CRU) of the Mayo Clinic. Administration of MDX-010 is performed by registered nurses and nurse practitioners in the CRU. By using the CRU, no charges are generated towards the study grant or the patient. Some patients continue to see their local physician for their clinical care requirements, which may save patients some money. In addition, the tertiary immunologic endpoints are now being studied in collaboration with Dr. James P. Allison at Memorial-Sloan-Kettering, and these costs are being defrayed by the laboratory budgets of Drs. Kwon and Allison.

VI. Informal Observations Pertaining to Prostate Cancer Patients Receiving AA Therapy Alone versus Combination AA + MDX-010 Treatment.

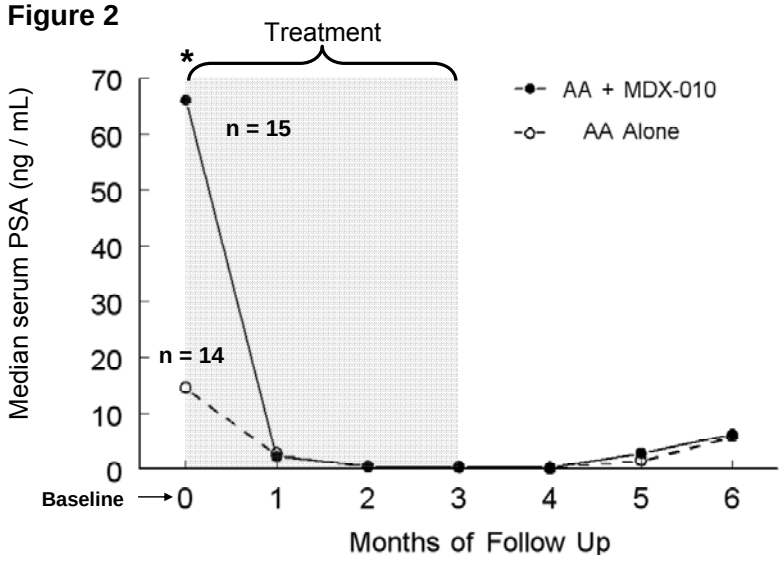
To date, we have enrolled a total of 46 prostate cancer patients onto study at the Mayo Clinic. Of these, 42 patients have been randomized to receive either initial AA (control) therapy alone or concurrent AA + MDX-010 treatment. Of the 42 patients who have been randomized to one of the two treatment arms of study, 29 patients now have sufficient follow-up to assess responses to protocol therapy including 14 control patients who have received AA therapy alone and 15 patients who have received concurrent AA + MDX-010 treatment.

In general, the demographics and histopathologic features of the first 38 prostate cancer patients (as of November 2007) randomized to the two arms of this study (of which 29 have adequate follow up to report) have been relatively balanced in terms of age, stage and Gleason Score as is described in **Table 1** (right). As indicated, > 50% of these patients have presented with advanced stage disease, and most have pretreatment biopsies revealing aggressive cancers encompassing Gleason Scores of 8 or greater.

TABLE 1	Arm A (N=20)	Arm B (N=18)
Age median (range)	65.5 (41.0-84.0)	63.0 (47.0-85.0)
Gleason Score Biopsy		
6	0	1 (5.56%)
7	5 (25%)	5 (27.78%)
8	3 (15%)	3 (16.67%)
9	11 (55%)	6 (33.33%)
10	1 (5%)	3 (16.67%)
T Stage		
T1	2 (10%)	0
T2	6 (30%)	5 (27.78%)
T3	7 (35%)	12 (66.67%)
T4	5 (25%)	1 (5.56%)
N Stage		
N0	11 (55%)	12 (66.67%)
N1	9 (45%)	6 (33.33%)
M Stage		
M0	12 (60%)	10 (55.56%)
M1	8 (40%)	8 (44.44%)
Disease Site		
Visceral	1 (5 %)	0
Bone	6 (30%)	4 (22.22%)
Soft Tissue	1 (5%)	2 (11.11%)
Other	9 (45%)	8 (44.44%)
Disease Status		
Measurable	3 (15%)	1 (5.56%)
Measurable and evaluable	7 (35%)	4 (22.22%)
Evaluable but not measurable	4 (20%)	7 (38.89%)
PSA only	6 (30%)	6 (33.33%)

Yet, despite the randomized design of this trial and the balanced profile of patient disease (stage and grade) as is depicted in **Table 1**, patients who have been randomized to receive initial AA therapy alone have presented with baseline PSA values (median 14.6 ng/mL) that are statistically lower than the PSA values (median 66 ng/mL) of those patients that have been randomized to receive concurrent AA therapy + MDX-010 infusion (**Figure 2**; *****p = 0.03). It is anticipated that this disparity in baseline PSA values between control and treatment groups will eventually rectify itself (to achieve parity in PSA values) as this study matures to its full accrual of patients. At present, the disparity in baseline PSA cannot be accounted for due to imbalanced accrual of patients defined by disease stage or histologic grade.

Figure 2



Nevertheless, another striking observation depicted in **Figure 2** is that median PSA values for patients who receive concurrent AA therapy + MDX-010 infusion decline to achieve parity with absolute PSA values for patients who receive AA therapy alone, even after only 1 month of study treatment (no differences in absolute, 1-6 month, median PSA values; all p values > 0.05).

Figure 3

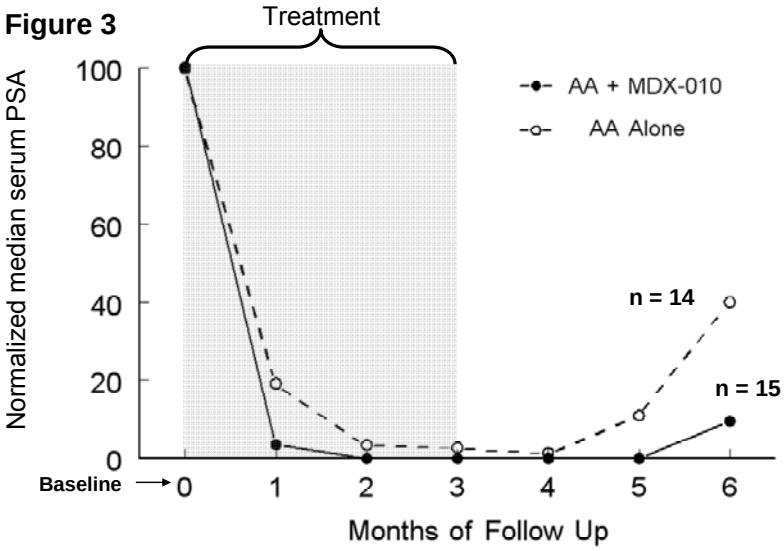


Figure 3 depicts the same data with PSA values normalized against baseline PSA values calculated as 100%. As is depicted in **Figure 3**, the percent-reduction in median PSA for patients who receive concurrent AA therapy + MDX-010 infusion is greater than the percent-reduction in median PSA for patients who receive AA therapy alone. Moreover, the percent-reduction in median PSA remains lower for AA therapy + MDX-010 treated subjects relative to control subjects even after cessation of AA therapy.

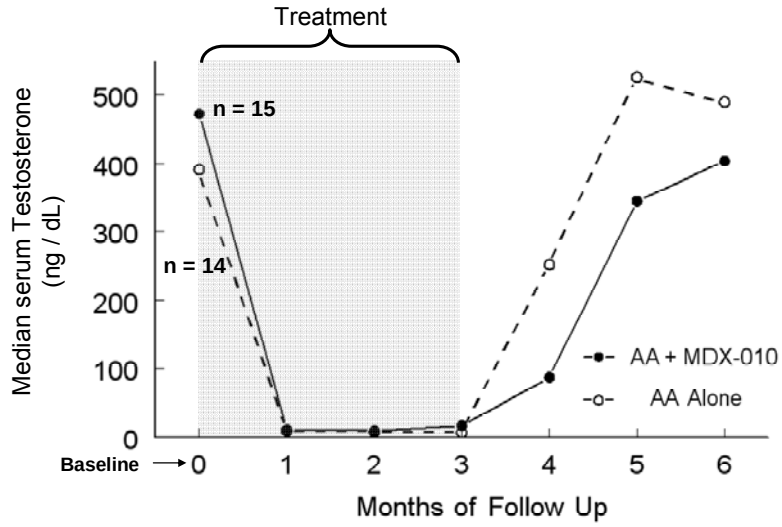


Figure 4 suggests that the increased reduction in serum PSA for the AA therapy + MDX-010 treated subjects (compared to AA alone treated control subjects) is not likely mediated by affecting serum testosterone production. However, it is worth noting that there is a trend towards delayed testosterone recovery in subjects treated with combined AA + MDX-010. Given that LHRH supra-agonists (i.e., Lupron) is a synthetic peptide and potential "antigen", and that MDX-010 potentiates responses against immunogenic targets (and sometimes induces autoimmune hypophysitis), it will be of great interest to assess whether this trend delineates itself further as the study matures. If so, one might conclude that combined LHRH supra-agonist + MDX-010 treatment might not be exclusively potentiating treatment responses via induction of prostate cancer-specific immune-mediated mechanisms.

If one takes into consideration that the pretreatment (baseline) PSA level acts as a determinant of PSA rebound after therapy (i.e. the higher the initial PSA, the more aggressively the PSA will rebound following cessation of therapy), one might interpret our data as supporting a benefit to combined AA + MDX-010 treatment when compared to AA therapy alone. However, further accrual of subjects, achieving parity in baseline PSA values for the two populations under study, will be required before firm conclusion can be reached. At the most superficial level, our current data indicates that combined AA + MDX-010 treatment may result in greater and more durable "secondary endpoint" PSA responses than is observed in patients receiving AA therapy alone. Our data also reveals novel information pertaining to interval until testosterone normalization following AA therapy.

In addition to the general findings described above, we believe it is worth mentioning several semi-remarkable responses that we have observed for individuals who have received combination AA + MDX-010 treatment. For instance, we have now treated at least three men who were referred to Mayo with rising PSAs despite initiation of hormone therapy (typically Casodex or Lupron alone). **Figure 5** depicts the PSA response for one such individual (41 year-old) who presented with severe urinary tract obstruction, widely metastatic prostate cancer and a rising PSA while on 2 weeks of Casodex anti-androgen therapy. Following randomization of this patient to the treatment

arm of study, this patient received Lupron as well as MDX-010 infusion. Within the first week after treatment, this patient's PSA declined from 168 ng / mL to 26 ng / mL. By four weeks after initiation of treatment, this individual's PSA was 2.6 ng / mL (12-19-07). Upon presentation, the patient was noted to have a massively enlarged prostate which he stated "felt like he was sitting on a softball". At present, the patient reports being asymptomatic and appears to be experiencing post-obstructive diuresis. In our estimation, this patient's response to combined AA + MDX-010 treatment (both in terms of PSA response and alleviation of urinary tract obstruction) is dramatic and highly atypical of responses induced by AA therapy alone. The response observed in this patient, however, is very representative of responses elicited in advanced prostate cancer patients who receive combined AA + MDX-010 treatment.

Figure 5

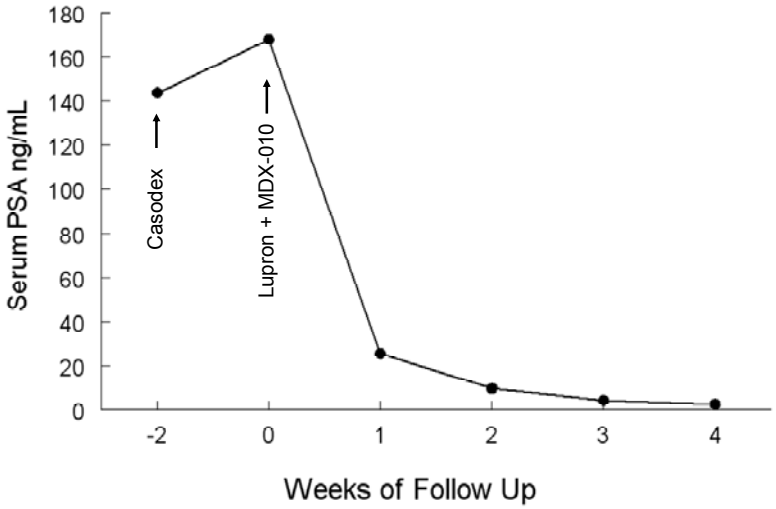


Figure 6

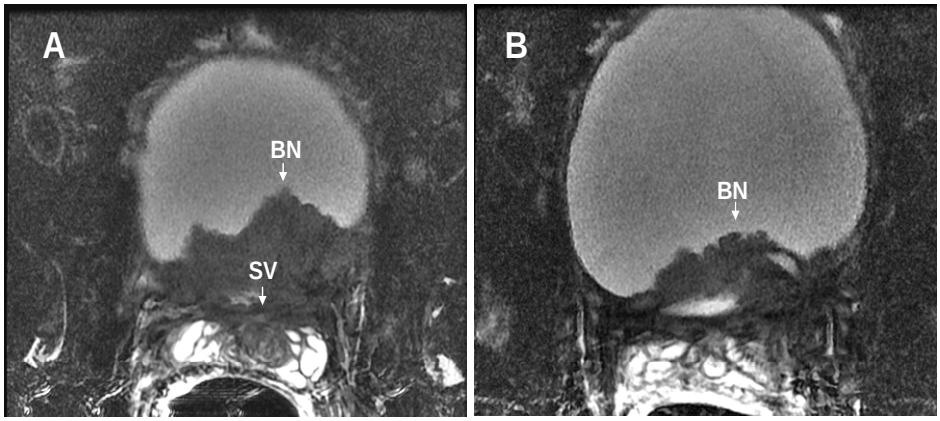


Figure 6 depicts a relatively common experience with AA therapy alone in one of the control patients accrued onto study. This patient initially presented with Gleason 10, stage cT4N0M0 adeno-carcinoma of the prostate with a serum PSA of 97.3 ng/mL. His baseline cystoscopy and MRI **(A)** confirmed extensive cancer invasion of the bladder neck (BN), seminal vesicles (SV) and perirectal fat (PF). The patient was randomized to receive only androgen ablative therapy (Lupron + Casodex). After 3 months of AA therapy, the patient's PSA was reduced to 72.6 ng/mL. At three months, repeat examination revealed persistent prostate cancer invasion of the bladder neck and obstructive uropathy as demonstrated by MRI and cystoscopy **(B)**. The patient was deemed a treatment failure and was taken off study.

Figure 7

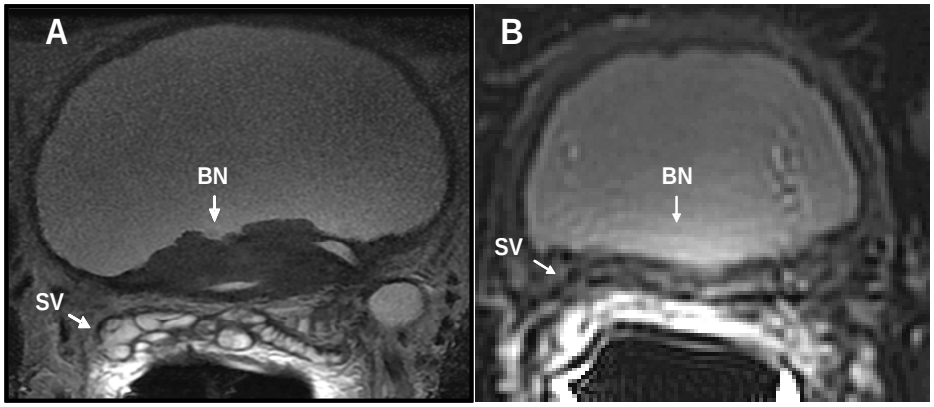


Figure 7 illustrates the experience with combination AA + MDX-010 treatment for one patient accrued onto study. This patient initially presented with Gleason 9, stage cT4N0M1 cancer of the prostate with a serum PSA of 12.3 ng/mL. Baseline MRI **(A)** and cystoscopy confirmed extensive carcinomatous invasion of the bladder neck (BN) and seminal vesicles (SV) with ureteral obstruction. The patient was in urinary retention and dependent upon catheter drainage. Three months after combination therapy, the patient's PSA was lowered to < 0.1 ng/mL (undetectable) and his prostate cancer invasion into the bladder neck was no longer evident on repeat MRI (B). In addition, the patient was able to void freely without a catheter by month 2 following treatment.

Figure 8

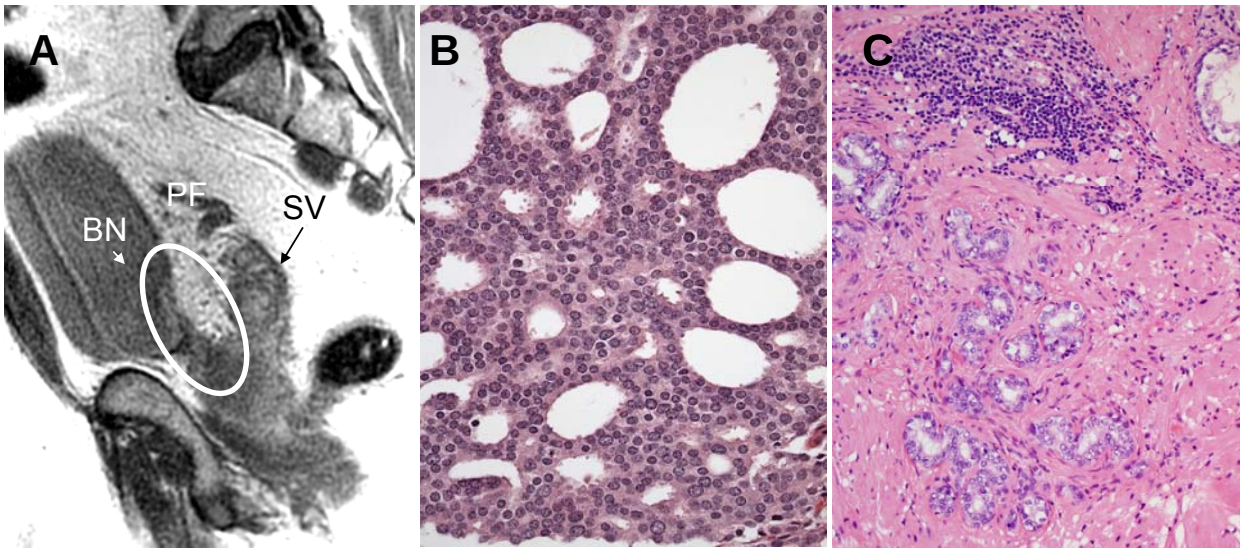
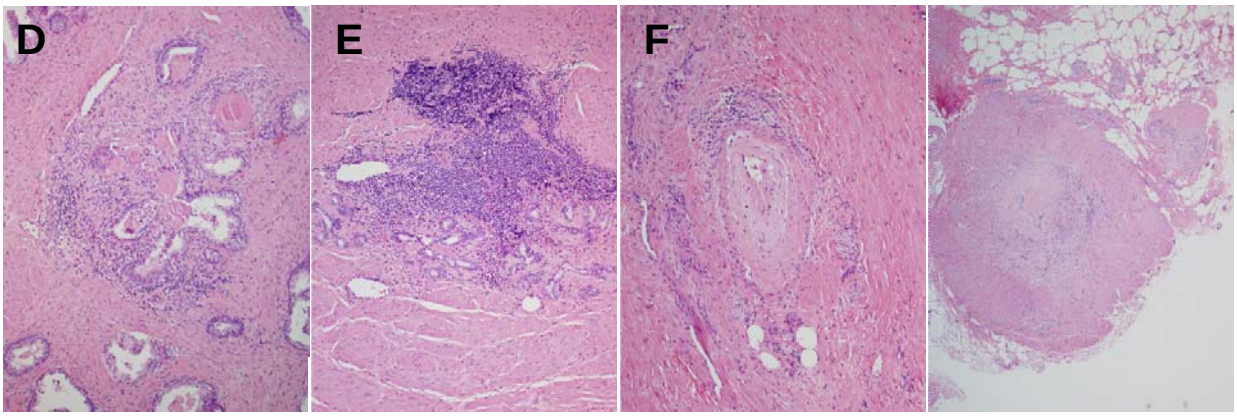


Figure 8 depicts a second patient who was treated with combination AA + MDX-010 therapy. This patient presented with Gleason 8, stage cT4N0M0 carcinoma of the prostate and serum PSA of 8.8 ng/mL. Baseline cystoscopy and MRI **(A)** confirmed extensive carcinomatous invasion of the bladder neck (BN), perivesical fat (PF) and seminal vesicles (SV). The patient was in urinary retention with a fixed prostate by DRE.

Pretreatment biopsy **(B)** revealed Gleason 10 cancer (by outside read), and bilateral Gleason 8 disease (involving up to 40-80% of the entire left lobe) according to Mayo Pathology. Based on initial workup, the patient was not considered to be a suitable candidate for RRP. After 3 months of combination AA + MDX-010 therapy, his PSA lowered to < 0.1 ng/mL, and prostate cancer invasion of the bladder neck was no longer evident on repeat MRI or by cytoscopy. DRE revealed a soft and mobile prostate. The patient opted to go off study in order to undergo radical surgery. The surgical site was notable for extensive inflammation which was initially interpreted as widespread carcinomatous spread. The final surgical specimen, however, revealed no extraprostatic cancer. In addition, only scattered and minute foci of Gleason 6 cancer were observed within the prostate, exhibiting both treatment effects and inflammation **(C)**. The remainder of the specimen revealed atrophic glands and inflammation **(D & E)**. Also, evidence of extensive and highly atypical myointimal hyperplasia **(F)** and vascular occlusion **(G)** was noted in the surgical specimen. The patient is currently 1 year post surgery, with an undetectable PSA.

Figure 8



Finally, a number of control patients have crossed over to receive MDX-010 infusion. A formal analysis of responses to MDX-010 treatment is currently pending as will be analyses of secondary responses for patients placed back on androgen ablative therapy.

VII. Summary of Adverse Events (AEs), Serious Adverse Events (SAEs), and Immune-related Adverse Events (irAEs)

On-Study AEs Related to Study Medication With At Least 5% Frequency – Treated Subjects (Medarex Data)

System Organ Class	Number of Subjects (%) - (N=1654)						
Preferred Term	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**	**Grade 5**	**Unknown**	**Ay Grade**
Any Adverse Experience	372 (22.5)	418 (25.3)	302 (18.3)	43 (2.6)	8 (0.5)	6 (0.4)	1149 (69.5)
General Disorders and Administration Site Conditions	361 (21.8)	230 (13.9)	51 (3.1)	4 (0.2)	2 (0.1)	3 (0.2)	651 (39.4)
Fatigue	230 (13.9)	137 (8.3)	27 (1.6)	2 (0.1)	0	0	396 (23.9)
Pyrexia	119 (7.2)	33 (2.0)	8 (0.5)	0	0	2 (0.1)	162 (9.8)
Injection Site Reaction	89 (5.4)	39 (2.4)	3 (0.2)	0	0	2 (0.1)	133 (8.0)
Gastrointestinal Disorders	330 (20.0)	175 (10.6)	132 (8.0)	11 (0.7)	1 (<0.1)	1 (<0.1)	650 (39.3)
Diarrhea	184 (11.1)	111 (6.7)	84 (5.1)	1 (<0.1)	0	0	380 (23.0)
Nausea	216 (13.1)	45 (2.7)	11 (0.7)	0	0	2 (0.1)	274 (16.6)
Abdominal Pain	87 (5.3)	41 (2.5)	14 (0.8)	0	0	0	142 (8.6)
Vomiting	78 (4.7)	34 (2.1)	13 (0.8)	0	0	0	125 (7.6)
Colitis	4 (0.2)	21 (1.3)	63 (3.8)	9 (0.5)	0	1 (<0.1)	98 (5.9)
Skin and Subcutaneous Tissue Disorders	374 (22.6)	196 (11.9)	44 (2.7)	2 (0.1)	0	0	616 (37.2)
Rash	195 (11.8)	118 (7.1)	27 (1.6)	1 (<0.1)	0	0	341 (20.6)
Pruritus	213 (12.9)	75 (4.5)	5 (0.3)	0	0	0	293 (17.7)
Metabolism and Nutrition Disorders	108 (6.5)	59 (3.6)	42 (2.5)	4 (0.2)	0	0	213 (12.9)
Anorexia	72 (4.4)	36 (2.2)	6 (0.4)	0	0	0	114 (6.9)
Nervous System Disorders	130 (7.9)	49 (3.0)	19 (1.1)	0	0	1 (<0.1)	199 (12.0)
Headache	75 (4.5)	31 (1.9)	8 (0.5)	0	0	0	114 (6.9)

On-Study AEs Related to Study Medication With At Least 5 Percent Frequency – Treated Subjects (Mayo Data)

System Organ Class	Number of Subjects (%) - (N=36)						
Preferred Term	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**	**Grade 5**	**Unknown**	**Any Grade**
Any Adverse Experience	17 (55%)	5 (16%)	2 (6%)	1 (3%)			25 (81%)
General Disorders, Admin. Site Conditions, Constitutional Symptoms	14 (45%)						14 (45%)
Fatigue	14 (45%)						14 (45%)
Constitutional Symptoms	2 (6%)						2 (6%)
Gastrointestinal Disorders	1 (3%)	2 (6%)	1 (3%)				4 (13%)
Diarrhea (no colostomy)	1 (3%)	1 (3%)	1 (3%)				3 (10%)
Skin and Subcutaneous Tissue Disorders	3 (10%)	2 (6%)					5 (16%)
Rash		2 (6%)					2 (6%)
Pruritus	2 (6%)	1 (3%)					3 (10%)
Metabolism and Nutrition Disorders	4 (13%)		2 (6%)				6 (19%)
Hyperkalemia	2 (6%)						2 (6%)
Lipase	1 (3%)		1 (3%)				2 (6%)
Nervous System Disorders	2 (6%)						2 (6%)
Neuro-motor	2 (6%)						2 (6%)
Hepatic	10 (32%)			1 (3%)			11 (35%)
SGOT (AST)	9 (29%)		1 (3%)				10 (32%)
Bilirubin	6 (19%)						6 (19%)
SGPT(ALT)	5 (16%)			1 (3%)			6 (19%)
Renal/Genitourinary	6 (19%)						6 (19%)
Pollakiuria	3 (10%)						3 (10%)
Creatinine	2 (6%)						2 (6%)
Endocrine	14 (45%)	1 (3%)					15 (48%)
Hot Flashes	14 (45%)	1 (3%)					15 (48%)

On the prior page, we report our overall experience with AEs and serious AEs, irrespective of cause, for patients treated in our study (bottom table). In addition we demonstrate that, in general, the AE and SAE profile that we have observed does not significantly differ from what is reported in the investigator brochure for MDX-010 provided by Medarex (upper table). In considering AEs and SAEs associated with our study, the following information is worth noting. We do report a slightly higher overall rate of AEs than is listed in the MDX-010 investigator brochure. Our higher overall rate of AEs is primarily due to increased rates of grade 1 events, mostly due to the reporting of hot flashes induced by hormone therapy. Also, one should note that the principle treatment-specific AE profile associated with MDX-010 primarily emanates from induction immunologic responses — the mechanism whereby MDX-010 effects treatment responses — including autoimmunity, which is further believed (by some) to be requisite in order to elicit regression of melanoma; primarily S.A. Rosenberg at NCI. As such, most of the treatment-relevant AEs associated with MDX-010 are immunologic in nature and, thus, termed either immune-related AEs (grade 1-2) or serious immune-related AEs (> grade 3). To date (see Tables below), the total Medarex experience with treatment of 1654 patients (most of whom have received **10 mg/kg** of MDX-010 **every 3 weeks** in combination with other agents) indicates an irAE rate of 51% (848 subjects) and a Serious irAE rate of 13% (214 subjects).

Immune-related AEs as of 31-Mar-2007 (Medarex Data)

Clinical Database	
Number of Subjects	1654
Number (%) of subjects with irAEs[a]	848 (51)
Number (%) of subjects with serious irAEs[a]	214 (13)

[a]Based on treatment-related adverse events retrieved from the clinical database using pre-defined MeDRA terms that were considered potential irAEs.

Immune-related AEs (Mayo Data)

Clinical Database	
Number of Subjects	36
Number (%) of subjects with irAEs[a]	8 (22)
Number (%) of subjects with serious irAEs[a]	3 (8.3)

With regards to our experience at Mayo encompassing the first 36 prostate cancer patients who have received a **single dose of MDX-010 at 3 mg/kg** (see Tables above), we have observed an irAE rate of 22% (8 patients) and a Serious irAE rate of 8.3% (3) patients. For those 8 study patients who have experienced grade 1-2 irAEs, the principle sources have been either asymptomatic (incidental) or transiently-observed rash, increases in liver (ALT or AST) or pancreatic function tests (lipase), diarrhea or self-resolving colitis found on routine colonoscopy performed for colon cancer screening. For those 3 study patients who have experienced grade 3-4 irAEs, principle sources have been symptomatic diarrhea associated with dehydration (1 patient), transient and self-resolving elevation of pancreatic enzymes (both lipase and amylase in 1 patient) and marked and symptomatic elevation of liver enzymes (ALT and AST). Interestingly, with regards to this latter patient (who encompasses the worst AE for this study having experienced grade 4 toxicity), onset of liver symptoms was extremely late relative to MDX-010 administration. Nevertheless, the patient was treated with the presumptive diagnosis of MDX-010 autoimmune hepatitis (with oral steroid) and his liver

functions rapidly normalized. Shortly thereafter, the patient (off study) was placed back on androgen-ablative (Casodex) therapy and suffered recurrence of liver dysfunction. Casodex was withdrawn, and the patient rapidly recovered. Given that oral androgen blockers are well described to also induce liver dysfunction, combined with this patient's prior history of toxic exposure as a farm implement spray painter, the PI believes this patient's history of liver abnormalities may be more consistent with pre-existent history of liver compromise + hormone therapy toxicities rather than toxicity induced by MDX-010. As such, the actual serious irAE rate for the present study may be closer to 5.5% (2 patients). For all patients on study with an AE, the most aggressive treatment of an AE has required use of a one-time dosing and taper of a steroid in combination with supportive or expectant care. Medarex has also documented a serious irAE rate of 7% (1 of 14 subjects) for prostate cancer patients treated with a single dose of 3.0 mg/kg MDX-010. As such, our experience parallels an outside study experience with 3 mg/kg single-dose treatment for prostate cancer.

VIII. Summary and Concluding Remarks

- To date, we have enrolled 46 patients onto study, nearly doubling our total accrual of the prior two years. We have also experienced a major surge in accrual activity which provides us with confidence that we can complete this study within 2 – 2.5 years. This represents a major accomplishment given that the first 3.5 years of this award were essentially dedicated to the drafting of our phase II protocol as well as a number of required administrative, legal and regulatory activities including procurement of local IRB, FDA and HSRRB approval. In addition, we believe that our accrual is remarkable, especially when one considers that this trial was originally scripted to be a two-site study.

- We have made, what we believe, are heroic efforts to drive and bolster accrual for this study and will continue to do so. This will likely include the eventual addition of Mayo Jacksonville and Scottsdale as additional sites to conduct this study. Regardless, we believe that this study has now attracted sufficient attention, both nationally as well as internationally, to attract advanced prostate cancer patients from around the world for possible entry into our study. Perhaps, most importantly, we believe that we have now finally changed the cultural "group think" of many urologists and oncologists who, at first, naturally tend to bristle against investigations that are truly novel, intrinsically risky, but also most likely to advance the treatment of prostate cancer.

- This study continues to be one of only a few studies that attempts to exploit the hormone sensitivity of prostate cancer in order to improve therapeutic outcomes for patients with advanced or recurrent prostate cancer. Moreover, treatment of prostate cancer patients with MDX-010 remains, without question, a cutting-edge immunotherapeutic approach to treat human malignancy.

- We have made every effort to conduct this trial in a highly cost-effective and fiscally-responsible manner. We have made arrangements to defray costs associated with assessing immunologic (tertiary) endpoints for this study; currently being performed in collaboration with Memorial Sloan Kettering Cancer Center with Dr. James P. Allison. Moreover, we are providing charge-free infusion, patient care and free MDX-010 to patients on this study. As such, these costs are not being charged against our grant. We are also in negotiations to have the Mayo Comprehensive Cancer Center defray all costs associated with opening accrual for this study at Mayo Jacksonville and Scottsdale. Finally, if needed, we will absorb additional costs for the study in order to meet our target accrual of patients for this study. In short, we are not asking for any additional funds from the DOD even though we believe that such funds would be most helpful to drive this study to its completion in a seamless and timely fashion.

- Our preliminary observations pertaining to the first 29 patients on study, who have at least 6 months of follow up (15 treatment patients and 14 control patients), suggests a potential advantage to combined AA + MDX-010 treatment over AA therapy alone

in terms of accelerating PSA reduction (and magnitude of reduction) as well as increasing the relative durability of reduction. These findings must be regarded as preliminary however, and will hopefully be more rigorously confirmed as this study matures and achieves balanced accrual of prostate cancer patients who initially present with comparable levels of baseline PSA. Our preliminary studies also suggest that combined AA + MDX-010 does not apparently affect initial testosterone production but may affect long term testosterone recovery in patients being treated with hormone therapy for prostate cancer.

- We illustrate a number of anecdotal cases in which we have observed atypical and dramatic responses to combined AA + MDX-010 therapy. Such observations include a rapid reduction of serum PSA in patients who were experiencing a rising PSA during treatment with anti-androgen or AA therapy. In addition, we report anecdotal cases of rapid resolution of urinary tract obstruction and dramatic down-staging of locally advanced prostate cancer (involving the bladder or outside of the prostate) in response to combined AA + MDX-010.

- We demonstrate that the irAE and irSAE rates associated with a single 3 mg/kg dose of MDX-010 for treatment of prostate cancer patients (receiving AA therapy) is lower than the irAE and irSAE rates of patients reported in the most recent investigator brochure for MDX-010 who typically receive 10 mg/kg doses of study medication every three weeks.

- We report that alterations in various eligibility criteria (some requested by members of the DOD HSRRB and our own Urology faculty) have dramatically improved our ability to accrue patients onto this study. We additionally report that we have formally screened 84 additional patients who did not qualify for entry into our study.

- We would like to point out that we have been extremely responsive to enormous pressures placed on us by the DOD and Medarex for rapid accrual of patients onto this study. However, we also recognize that such pressures to facilitate rapid accrual of patients must be judiciously considered and implemented in such a fashion to avoid contamination of the clinical and scientific integrity of our study. As such, we believe the changes we have instituted thus far represent a most reasonable compromise. Finally, we would like to point out that this entire process has been a valuable learning experience for the DOD, our institution and the study PI. We understand that this learning experience has already enhanced the DOD's approach to funding important future clinical trials for the treatment of cancer.

- As an aside, we would like to point out that some patient tissues obtained on this study were used to demonstrate that a T cell-inhibitory ligand, B7-H3, is retained by AA-treated prostate cancer tumor cells (DOD Award DAMD17-02-1-0245 acknowledged, in Roth et al, Cancer Res. 2007 Aug 15;67[16]:7893-900). Increased expression of B7-H3 by untreated prostate cancer tumor cells portends an especially poor prognosis for patients with prostate cancer even after RRP. This observation has now been externally independently validated by Dr. James P. Allison's group at

MSKCC, (B7-H3 and B7x are highly expressed in human prostate cancer and associated with disease spread and poor outcome. Proc Natl Acad Sci U S A. 2007 Dec 4;104(49):19458-63). Thus, B7-H3 encompasses a novel inhibitor of host immunity and may partially explain the refractory nature of prostate cancer in response to immunotherapy. Conversely, B7-H3 might easily be targeted for antitumoral immunotherapy. Given that B7-H3 is likely retained by AA-treated as well as androgen-refractory and metastatic prostate tumor cells (a larger study confirming this has now been completed by our group), B7-H3 encompasses a promising target to improve prostate cancer treatment overall.

- In summary, we are requesting a 2.5 year no-cost extension for our award to continue with our study "A Phase II Immunotherapeutic Trial: Combination Androgen Ablative Therapy and CTLA-4 Blockade as a Treatment for Advanced Prostate Cancer". We believe that this is an entirely reasonable and highly ethical request to continue to support meritorious work that will likely prove important to enhance advanced prostate cancer treatment for the future.

References

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Appendices

Not applicable.